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A basis for accelerated progression of diabetic nephropathy in Pima Indians

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A basis for accelerated progression of diabetic nephropathy in Pima Indians. The Pima Indians of Arizona not only have a much higher incidence of nephropathy due to type 2 diabetes than Caucasians, but they also lose their renal function at an accelerated rate after they develop diabetic nephropathy. This rapid loss of renal function occurs despite a younger age of onset of nephropathy and lower blood pressures and lipid levels, all of which would seem to predict a slower rate of progression of nephropathy. These findings suggest that other factors contribute to the rapid progression of renal disease in this population. In particular, glomerulomegaly in this population may contribute to the high rate of glomerular filtration rate (GFR) loss during the terminal, clinically manifest phase of nephropathy, because of the greater incremental loss of single-nephron GFR (SNGFR) with each nephron lost to sclerosis. In nine Pima Indians with type 2 diabetic nephropathy who underwent renal biopsy followed by serial iothalamate clearances for up to ten years, we examined the relationship between glomerular tuft volume at initial biopsy and the rate of GFR loss during the terminal phase. By multivariate analysis, significant independent effects of both glomerular volume ($P = 0.006$) and podocyte density ($P = 0.043$) were evident in these individuals. The effect of glomerular volume may result from a greater loss of intrinsic filtration capacity with each glomerulus lost, while the effect of podocyte density may reflect the destabilizing influence of "podocyte insufficiency" on the glomerular tuft. Similar factors may play a role in the rapid loss of GFR associated with progressive glomerular diseases in other indigenous populations in whom glomerulomegaly and glomerulopenia coexist.

The natural history and pathophysiology of diabetic nephropathy have been studied in Pima Indians for well over a decade [1–4]. This population group has a very high incidence of end-stage renal disease (ESRD), more than 20 times that of the general US population, related to type 2 diabetes and diabetic nephropathy. Unlike European populations with type 2 diabetes and kidney disease, renal involvement occurs relatively early in life with the Pima Indians and the significant incidence of non-diabetic kidney disease seen in older European populations with type 2 diabetes is not observed. The Pima population is relatively small, so the incidence of less common kidney diseases cannot be precisely determined, although these diseases probably account for <5% of all ESRD among Pima Indians [3].

The development and progression of diabetic nephropathy—from the microalbuminuric incipient stage, through the macroalbuminuric overt stage on to ESRD—has been studied in a large number of individuals using both functional and structural techniques [1, 4, 5]. In these subjects, the glomerular filtration rate (GFR) as determined by urinary iothalamate clearance has been measured serially for periods of up to 10 years. In additional studies, the physiologic and structural determinants of GFR also were measured. A relative stability of GFR early on in subjects initially presenting with microalbuminuria has been demonstrated, while subjects with macroalbuminuria at the time of enrollment have been found to have a much more rapid decline in their GFR (35%) over five years of follow-up [2]. It is notable that the macroalbuminuric subjects had "normal" values of GFR at study entry (124 ± 7 mL/min), comparable to Pima Indians with normal glucose tolerance. This apparent normality must be interpreted critically, however, inasmuch as in the 1997 study of Pagtalunan and colleagues, a macroalbuminuric group of diabetic Pima Indians with only a slightly lower GFR (103 ± 12 mL/min) had an incidence of global glomerular sclerosis that was much higher (19 ± 4%) than that in subjects with diabetes of short duration (3 ± 1%) [4]. This highlights the difficulty of assessing renal disease progression during the clinically silent "pre-slippery slope" phase. Physiologic, compensatory hyperfunction in the remnant nephrons can mask significant losses of intrinsic filtration capacity, and thus obscure the initial structural phase of disease progression.

The principal aim of this report is to address one specific question: Do the Pima Indians have a faster rate of progression of renal disease in type 2 diabetes than comparison non-indigenous populations and, if so, why? This is different from the question of whether or not there is a higher incidence of ESRD with type 2 diabetes

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in this population: the answer to that question is easier. There is about a 14-fold increased risk of ESRD in the Pima Indian population. Understanding whether and why the rate of progression to ESRD is faster in Pima Indians may have significance in explaining the alarming epidemics of renal disease occurring in indigenous populations around the world, inasmuch as the Pima Indians appear to share some characteristics with these other population groups.

REVIEW OF PERTINENT RESULTS

The question of whether Pima Indians with type 2 diabetic nephropathy have a faster rate of progression of renal function is in fact ambiguous until the phrase "rate of progression" is properly defined. The most unambiguous definition would probably be based on the time from the onset of diabetes mellitus to the development of ESRD. Although this type of data is available in the Pima Indian population, it is absent from the traditional comparison population of Caucasian Europeans. Clearly, another approach to defining the rate of progression is necessary.

This is made challenging by the fact that the concept of progression is multifaceted, including both structural and functional aspects. Even accepting that "progression" can mean variously development of irreversible structural lesions, increases in urinary albumin excretion rates or loss of GFR does not resolve the issue, as the latter two physiologic variables are often quite non-monotonic. This is the result of the fact that they are the product of labile physiologic factors superimposed on more slowly developing, largely irreversible structural lesions. Thus, patients may have urinary albumin excretion rates in and out of the abnormal range for years before becoming persistently albuminuric. In our longitudinal study of the development of overt nephropathy in initially microalbuminuric Pima Indians [1], for example, there was a three to four year difference in the median time for progression from microalbuminuria to macroalbuminuria, depending upon whether this was defined as "first" progression to macroalbuminuria [the first time a urinary albumin/creatinine (A/C) ratio over 299 mg/g was found] or "final" progression (the time from which all subsequent A/C ratios were over 299 mg/g).

With respect to progression from microalbuminuria to macroalbuminuria, it is possible to compare this rate in Pima Indians with that in predominantly Caucasian hypertensive patients described in a recent study of Parving et al [6]. The placebo group (N = 201) in their treatment study started out with a urinary albumin excretion (UAE) rate of about 55 μg/min, quite comparable to our microalbuminuric group's initial spot albumin/creatinine (A/C) ratio of 81 mg/g [1]. At the end of 24 months of follow-up, about 15% of their patients had progressed to overt nephropathy (UAE >199 μg/min), whereas in our patient cohort about 10% of the 43 initially microalbuminuric patients had undergone "final" progression to macroalbuminuria [1]. Thus, with regard to this aspect of their disease, the Pima population does not seem to progress more rapidly, although the comparison population in this case is notable for being hypertensive.

Comparing rates of GFR loss between different populations is made difficult by the fact that quite different methods of estimating GFR have been used in the various studies. Our cohorts of Pima Indians were studied using urinary iothalamate clearances, while in other studies plasma clearances of ethylenediaminetetraacetic acid (EDTA) [7] or iohexol, creatinine clearance or doubling of serum creatinine was used. In addition, it is important to know from which starting point a loss of GFR is to be measured. Although the time of onset of diabetes is a logical starting point, in most populations, type 2 diabetes is discovered incidentally while investigating diabetic complications such as cardiovascular disease, making it difficult to estimate the date of onset of diabetes. Unlike other populations, because of universal biennial screening, the actual time of onset of impaired glucose tolerance and diabetes in the Pima Indians is generally known to within about two years.

In the study of Nosadini et al, over a 3 year follow-up period the average rate of loss of GFR in 34 macroalbuminuric subjects (UAE > 199 μg/min) was 3 mL/min/1.73 m^2 body surface area (BSA) per year, with half of these patients manifesting a decrease in GFR and half not [7]. Among those whose disease progressed, the average GFR loss was 11 mL/min/1.73 m^2/year. In the study of Christensen and colleagues of 34 macroalbuminuric type 2 diabetic patients with a median follow-up of 4 years, the mean rate of decrease in GFR was 5.3 mL/min/1.73 m^2 per year [8]. In our group of 34 Pima Indians with type 2 diabetes and macroalbuminuria at enrollment, there was a mean rate of loss of absolute GFR of 16 mL/min per year (unpublished data). Allowing for a median body surface area of 2.0, the average rate of GFR loss would be about 13.8 mL/min/1.73 m^2 per year, over four times the rate of decline of the patients in Nosadini's study and more than twice the rate of Christensen's patients. In the study of Nosadini and colleagues, about 18% of the proteinuric patients had progressed to ESRD by the end of follow-up, while none were reported to have progressed to ESRD during follow-up in Christensen's group of patients. In our long-term follow-up of Pima Indian subjects presenting with macroalbuminuria, on the other hand, the majority of the 35 subjects have progressed to ESRD by 10 years of follow-up.

The development of glomerular sclerosis rarely has been quantitatively studied. In Pima Indians there is a

trend for an increased frequency of sclerosis in subjects with more advanced stages of diabetic nephropathy [4]. In a longitudinal study of 12 Pima Indians initially presenting with microalbuminuria, there was a tendency ($P = 0.15$) toward an increase in the incidence of global sclerosis from 4% to 8% over four years of follow-up [5]. No comparable studies are available in comparison populations of type 2 diabetics. The high degree of uncertainty in determinations of the incidence of glomerular sclerosis from biopsy specimens makes estimation of the rate of development of global sclerosis problematic. This is compounded by the fact that sclerotic glomeruli are probably eventually resorbed, so that this index of progression is not cumulative.

What are the factors that may account for a faster rate of functional progression in Pima Indians with type 2 diabetic nephropathy? As a first approach it is useful to examine those characteristics in which Pima Indians differ from comparison populations. Any of the following might be responsible for their propensity for more rapid progression: increased glomerular size [4, 10], decreased glomerular epithelial cell density [4], early onset of diabetes, and poor glycemic control [1, 6, 7]. On the other hand, other factors exist that might be expected to mitigate the risk of rapid progression such as a lack of early hypertension [1, 6, 7] and a lack of hyperlipidemia.

In the considerations that follow, only aspects of glomerular structure, including glomerular size and podocyte number, will be addressed. Schmidt and colleagues showed some time ago that the mean glomerular volume of both diabetic and non-diabetic Pima Indians under 60 years of age (4.1×10^6 μm^3) was greater than that of both whites (2.3×10^6 μm^3) and blacks (about 3.2×10^6 μm^3) [9]. In the study of Pagtalunan et al, the average glomerular volume in newly diagnosed diabetic Pima Indians with normal GFR was about twice that of the predominantly Caucasian kidney transplant donors who constituted the control population (5.4×10^6 μm^3 vs. 2.6×10^6 μm^3) [4].

Since the average GFR is not higher in non-diabetic Pima Indians than in the comparison groups, it is likely that either the single-nephron ultrafiltration coefficient for the Pima population is unusually low for their large glomerular size; the net pressure for ultrafiltration is low; or the single-nephron GFR is in fact elevated and the total number of nephrons is diminished. With regard to the first possibility, Østerby and colleagues have shown a strong relationship between filtration surface area and GFR in patients with long-term type 1 diabetes with albuminuria [10]. We have calculated an estimated single-nephron ultrafiltration coefficient (SNK_f) using the hydrodynamic model of Drumond and Deen [11] for Pima Indians with early onset diabetes, normal renal function and normoalbuminuria [5]. The morphometrically determined SNK_f was approximately twice as great as that in controls calculated using the same model [12]: 15 versus 7 nL/min/mm Hg. With regard to the net intraglomerular pressures sustaining ultrafiltration, both plasma oncotic pressure and mean arterial pressure (as a surrogate for intraglomerular hydrostatic pressure) are indistinguishable in Pima Indians from healthy controls from non-Indian populations, making a difference in the driving forces unlikely to lead to a significant difference in the net ultrafiltration pressure. Although the situation with respect to filtration pressure equilibrium is in general not well understood in humans, the fact that non-diabetic Pima Indians have the same filtration fraction (about 19%) as non-Pima subjects is consistent with their being in the same state of filtration pressure equilibrium as the latter.

The preceding considerations suggest that normal glomerular number is low even in healthy, non-diabetic Pima Indians, compared to Caucasian populations. Under the assumption that the average GFR is constant among all human populations (at least when normalized for body size or lean body mass), a low glomerular number would be a virtual corollary of the existence of an elevated SNGFR. Given that the SNK_f of Pima Indians is twice that of comparison populations and that the driving forces for ultrafiltration are equal, the SNGFR in the Pima group should be on average about twice that of the comparison group, and the number of nephrons in the former about half that in the comparison group.

We and others [10] have shown that in a number of progressive glomerular diseases the best correlate for loss of GFR in the latter stages is the development of global glomerular sclerosis. Assuming an equal rate of development of glomerular sclerosis, the increased single-nephron GFR in Pima Indians implies that for each glomerulus lost a greater decrement in the overall GFR would occur. In particular, once the slippery slope has been reached, the rate of functional loss in Pima Indians (diabetic or not) would be about two times that of comparison populations for the same rate of glomerular loss. This factor alone could account for much of the difference in rates of GFR loss described above.

Little direct experimental data are available to address this possibility. We have made serial determinations of GFR by iothalamate clearance, however, in a large cohort of Pima Indians with type 2 diabetes who were found to have macroalbuminuria at screening. Ten of these subjects had kidney biopsies performed at the beginning of their follow-up [4]. Of these, nine had sufficiently linear GFR courses in their terminal phase to estimate rates of GFR loss (Fig. 1). Over 79 to 129 months of follow-up, these nine individuals lost GFR at a rate of about 18 mL/min/year, quite similar to the 16 mL/min/year in the group of 34 subjects as a whole. ESRD occurred in seven of the nine subjects during follow-up.

The relationship between the rate of GFR loss and

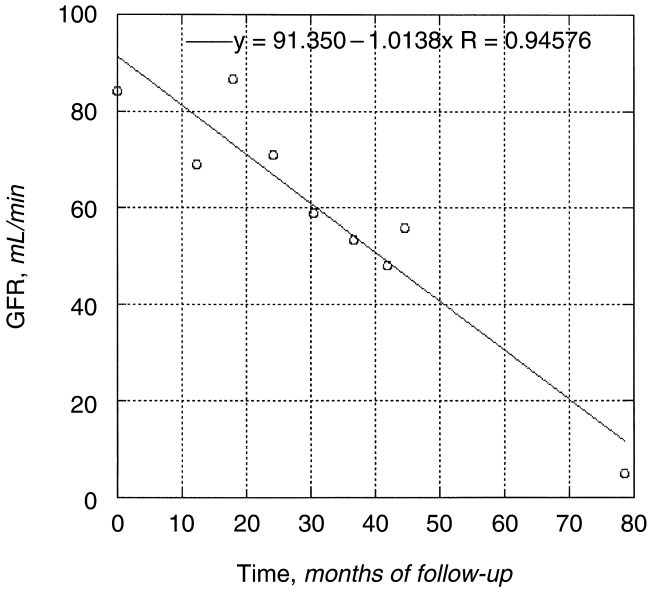

Fig. 1. Course of glomerular filtration rate (GFR; iothalamate clearance) over 80 months of follow-up in a Pima Indian with type 2 diabetes and macroalbuminuria at screening. The decline in the slope is linear.

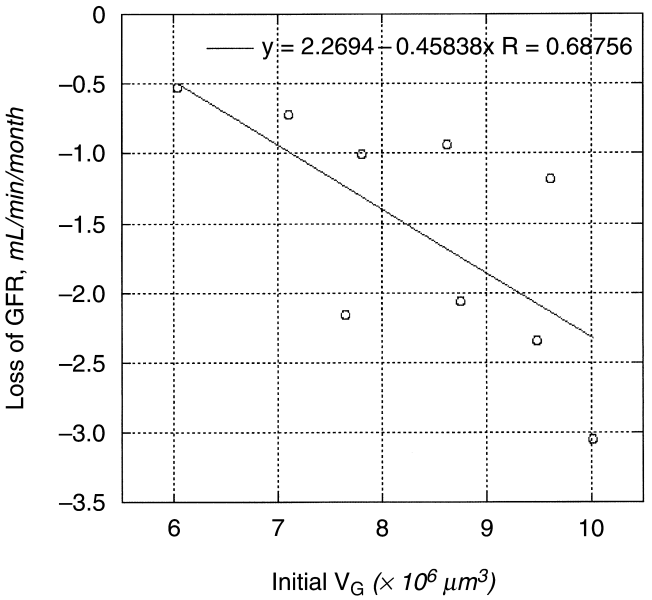

Fig. 2. Relationship between rate of loss of GFR (mL/min/month) and average glomerular volume (V_G) determined from the biopsy at study entry. The regression is significant ($P = 0.041$).

glomerular tuft volume was examined by linear regression. There was a significantly greater rate of GFR loss with increasing glomerular volume ($P = 0.041$; Fig. 2). About half of the variation in the rate of GFR loss could be explained by variation in glomerular volume ($r^2 = 0.47$). Because decreased podocyte density has been suggested to contribute to the development of glomerular sclerosis [4, 13] and podocyte density would be expected to decrease with increasing glomerular volume, it could be argued that this relationship just represents the contribution of glomerulomegaly to low podocyte densities. Therefore, the relationship between the rate of GFR loss and several possible explanatory variables was examined by multilinear regression (stepwise procedure), with the regression being performed on glomerular volume, initial GFR, incidence of global sclerosis, and either podocyte density or absolute podocyte number. If the effect of increased glomerular volume on GFR loss were mediated by decreases in podocyte density, the former variable should fall out of the multivariate regression. In fact, glomerular volume remained the strongest factor in the model, whether podocyte number or podocyte density also was included. The *t* value for glomerular volume actually increased with the addition of either of these variables (Table 1), suggesting that glomerular volume has an independent impact on the rate of GFR loss.

Table 1. Multilinear regression model for rate of GFR loss

Factor	*t* value	*P* value
Podocyte number		
V_G	−2.751	0.033
N_P	−2.634	0.039
Podocyte density		
V_G	−4.168	0.006
$N_{V,podo}$	−2.563	0.043

Abbreviations are: V_G, glomerular tuft volume; N_P, absolute podocyte number/glomerulus; $N_{V,podo}$, podocyte density − podocytes/tuft volume

CONCLUSION

The rate of loss of GFR in macroalbuminuric, type 2 diabetic Pima Indians appears to be greater than that in comparison Caucasian populations. Glomerulomegaly (probably associated with inherited glomerulopenia) could account for about a twofold greater rate of GFR loss in the clinically overt stage of nephropathy simply on the basis of a twofold greater loss of SNGFR with each nephron lost to glomerular sclerosis. Since other indigenous populations also have been suggested to have glomerulomegaly [14], it is possible that an increase in the terminal rate of GFR loss in these populations might occur independently of any greater rate of loss of nephrons due to the disease process than in comparison populations.

Of course, glomerulomegaly could contribute also to an increased rate of development of glomerular sclerosis by virtue of leading to a decreased podocyte density within the glomerulus. Under conditions of podocyte "sufficiency," a greater glomerular volume (and ultrafiltration capacity) in fact could be renoprotective, as glomerular hypertrophy may allow for adaptive rises in SNGFR with lesser increases in intraglomerular capillary

pressures [15]. The apparent equality of total podocyte number per glomerulus in different population groups [4], however, suggests that an inherent tendency toward podocyte insufficiency may be characteristic of populations with baseline glomerulomegaly.

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REFERENCES

1. Lemley KV, Blouch K, Abdullah I, *et al*: Glomerular permselectivity at the onset of nephropathy in type 2 diabetes mellitus. *J Am Soc Nephrol* 11:2095–2105, 2000
2. Nelson RG, Bennett PH, Beck GJ, *et al*: Development and progression of renal disease in Pima Indians with non-insulin-dependent diabetes mellitus. Diabetic Renal Disease Study Group. *N Engl J Med* 335:1636–1642, 1996
3. Nelson RG, Newman JM, Knowler WC, *et al*: Incidence of end-stage renal disease in type 2 (non-insulin-dependent) diabetes mellitus in Pima Indians. *Diabetologia* 31:730–736, 1988
4. Pagtalunan ME, Miller PL, Jumping-Eagle S, *et al*: Podocyte loss and progressive glomerular injury in type II diabetes. *J Clin Invest* 99:342–348, 1997
5. Lemley KV, Abdullah I, Myers BD, *et al*: Evolution of incipient nephropathy in type 2 diabetes mellitus. *Kidney Int* 58:1228–1237, 2000
6. Parving HH, Lehnert H, Brochner-Mortensen J, *et al*: The effect of irbesartan on the development of diabetic nephropathy in patients with type 2 diabetes. *N Engl J Med* 345:870–878, 2001
7. Nosadini R, Velussi M, Brocco E, *et al*: Course of renal function in type 2 diabetic patients with abnormalities of albumin excretion rate. *Diabetes* 49:476–484, 2000
8. Christensen PK, Larsen S, Horn T, *et al*: Renal function and structure in albuminuric type 2 diabetic patients without retinopathy. *Nephrol Dial Transplant* 16:2337–2347, 2001
9. Schmidt K, Pesce C, Liu Q, *et al*: Large glomerular size in Pima Indians: lack of change with diabetic nephropathy. *J Am Soc Nephrol* 3:229–235, 1992
10. Osterby R, Parving HH, Nyberg G, *et al*: A strong correlation between glomerular filtration rate and filtration surface in diabetic nephropathy. *Diabetologia* 31:265–270, 1988
11. Drumond MC, Deen WM: Structural determinants of glomerular hydraulic permeability. *Am J Physiol* 266:F1–F12, 1994
12. Squarer A, Lemley KV, Ambalavanan S, *et al*: Mechanisms of progressive glomerular injury in membranous nephropathy. *J Am Soc Nephrol* 9:1389–1398, 1998
13. Fries JW, Sandstrom DJ, Meyer TW, Rennke HG: Glomerular hypertrophy and epithelial cell injury modulate progressive glomerulosclerosis in the rat. *Lab Invest* 60:205–218, 1989
14. Young RJ, Hoy WE, Kincaid-Smith P, *et al*: Glomerular size and glomerulosclerosis in Australian aborigines. *Am J Kidney Dis* 36:481–489, 2000
15. Tenschert S, Elger M, Lemley KV: Glomerular hypertrophy after subtotal nephrectomy: Relationship to early glomerular injury. *Virchows Arch* 426:509–517, 1995